WER 6 OF 10 CAPLUS COPYRIGHT 2002 ACS
AN 1971:474117 CAPLUS
DN 75:74117
TI Site-directed cross-linking. A new approach to mapping antibody combining sites
AU Hadler, Nortin M.; Metzger, Henry
CS Natl. Inst. Arthritis Metab. Dis., Natl. Inst. Health, Bethesda, Md., USA
SO Proc. Nat. Acad. Sci. U. S. (1971), 68(7), 1421-4
CODEN: PNASA6
DT Journal
LA English
AB The .gamma.A myeloma **protein** 315 from the mouse was affinity-**labeled** with m-nitrobenzenediazonium fluoroborate which leads to selective modification of the tyrosine at position 34 in the light chains of this **protein**. The azotyrosine bound was reduced with dithionite to form **3-aminotyrosine**. The aryl amino group of the aminotyrosine was selectively reacted with the bifunctional reagent 1,5-difluoro-2,4-dinitrobenzene. Cross links were formed between the aminotyrosine and at least 2 residues-one on the same light chain and one in the Fd region of the heavy chain

4041300 EMBASE
DN 1974041300
TI The modification of affinity labeled antibodies.
AU Hadler N.; Metzger H.
CS Arthrit. Rheum. Branch, Nat. Inst. Arthrit. Metab. Digest. Dis., NIH, Bethesda, Md. 20014, United States
SO Molecular Immunology, (1973) 10/7 (455-460).
CODEN: IMCHAZ
DT Journal
FS 026 Immunology, Serology and Transplantation
025 Hematology
LA English
AB **Antibodies** which have been affinity **labeled** at a tyrosyl residue with a diazonium **labeling** reagent could be further modified. By reductively cleaving the azotyrosine bond, **3 aminotyrosine** (3 NH2Tyr) was generated in situ, and this residue could then serve as an initiation point for further modifications, for example with the bifunctional reagent difluoro dinitrobenzene (F2DNB). The affinity **labeled** nitrophenyl binding **protein** of the plasma cell tumor MOPC 315 was easily reduced with small amounts of dithionite (Na2S2O4). When F2DNB was added it reacted initially at the 3 NH2Tyr at position 34 on the light chain and then subsequently with other amino acid residues leading to the formation of light heavy and intra light chain cross links. Although under suitable conditions F2DNB also reacted with the native **protein** of MOPC 315, the reaction occurred largely on the heavy chain. These results testify to the high specificity obtainable using these procedures. Experiments involving the reaction of F2DNB with either **labeled** unreduced **protein** , or **labeled** reduced **protein** in the presence of the ligand dinitrophenyl aminocaproate, suggests that the F2DNB may in part have functioned as an affinity **labeling** reagent. Parallel studies were performed with the phosphorylcholine binding **protein** of plasma cell tumor TEPC 15. This **protein** affinity **labeled** at two alternative tyrosines on the light chains. The azo bonds were much more difficult to reduce with dithion

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5 ANSWER 1 OF 2 CAPLUS COPYRIGHT 2002 ACS
AN 1973:544769 CAPLUS
DN 79:144769
TI Modification of affinity-labeled antibodies
AU Hadler, North; Metzger, Henry
CS Natl. Inst. Arthritis, Metab. Dig. Dis., Natl. Inst. Health, Bethesda, Md., USA
SO Immunochemistry (1973), 10(7), 455-60
CODEN: IMCHAZ
DT Journal
LA English
AB **Antibodies** which had been affinity **labeled** at a tyrosyl residue with a diazonium **labeling** reagent could be further modified **usefully**. By reductively cleaving the azotyrosine bond, **3-aminotyrosine** (3-NH2Tyr) was generated in situ, and this residue could then serve as an initiation point for further modifications, e.g., with the bifunctional reagent difluoro-dinitrobenzene (F2DNB). The affinity **labeled** nitrophenyl binding **protein** of the plasma cell tumor MOPC 315 was easily reduced with small amts. of Na2S2O4. When F2DNB was added, it reacted initially at the 3-NH2Tyr at position 34 on the light chain and then subsequently with other amino acid residues leading to the formation of light-heavy and intra-light chain cross-links. Although under suitable conditions F2DNB also reacted with the native **protein** of MOPC 315, the reaction occurred largely on the heavy chain. Expts. involving the reaction of F2DNB with either **labeled**-unreduced **protein**, or **labeled**-reduced **protein** in the presence of the ligand dinitrophenyl-aminocaproate, suggested that the F2DNB may in part have functioned as an affinity **labeling** reagent. Parallel studies were performed with the phosphorylcholine binding **protein** of plasma cell tumor TEPC 15. This **protein** affinity **labeled** at 2 alternative tyrosines on the light chains. The azo bonds were much more difficult to reduce with dithionite. When they were cleaved, some of the sites remained completely inactive, whereas others bound normally. The stoichiometry suggests that those sites with a 3-NH2Tyr at position 92 were inactivated while those with 3-NH2Tyr at position 34 were unaffected. Native **protein**-15 reacted only minimally with F2DNB under the conditions used whereas the **labeled**-reduced **protein** became modified on the light chains exclusively.

L5 ANSWER 2 OF 2 USPATFULL
AN 2002:29249 USPATFULL
TI Detection of binding reactions using labels detected by mediated catalytic electrochemistry
IN Stewart, David H., Durham, NC, United States
Groelke, John W., Raleigh, NC, United States
Thorp, H. Holden, Carrboro, NC, United States
Eckhardt, Allen E., Durham, NC, United States
PA Xanthon, Inc., Research Triangle Park, NC, United States (U.S. corporation)
The University of North Carolina at Chapel Hill, Chapel Hill, NC, United States (U.S. corporation)
PI US 6346387 B1 20020212
AI US 2000-722065 20001124 (9)
RLI Continuation-in-part of Ser. No. US 2000-603217, filed on 26 Jun 2000
Division of Ser. No. US 1998-179665, filed on 27 Oct 1998, now patented,

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(FILE 'HOME' ENTERED AT 08:22:13 ON 11 MAR 2002)

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, USPATFULL' ENTERED AT 08:22:44 ON
     11 MAR 2002
L1        371714 S (PROTEIN OR ANTIBOD?) (P) LABEL?
L2            78 S L1 (P)(5(W)HYDROXYTRYPTOPHAN)
L3            10 S L1 (P)(3(W)AMINOTYROSINE)
L4             3 S L2 (P)(ADVANTAG? OR USEFUL?)
L5             2 S L3 (P)(ADVANTAG? OR USEFUL?)

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